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"BACTERIOSIS" OF CARNATIONS.[1]

ALBERT F. WOODS.

DURING the past two or three years the writer has had under investigation a disease of the Bermuda, or Easter lily. Early in the course of this work it was observed that the disease of the lily possessed certain characteristics in common with one affecting carnations, first described by Dr. J. C. Arthur in a paper[2] read at a meeting of this association at Toronto, August 1889. Through this paper and the publications of Dr. Arthur[3] and of Arthur and Bolley,[4] the disease has come to be generally known as "bacteriosis" of carnations. In connection with the work on the lily disease,[5] "bacteriosis" of carnations has received considerable attention during the past year, and the object of this paper is to review briefly some of the results obtained.

For a general description of the disease the following, quoted from the bulletin by Arthur and Bolley already cited, covers the ground thoroughly:

> Bacteriosis is a disease of the carnation leaf, rarely attacking the stem or other parts of the plant. It generally starts in the leaf when immature, and is best diagnosed in the younger but full-sized leaves nearest the upper end of the stem. Taking such a leaf, which on its surface presents no unusual appearance to the eye, and holding it toward strong light, small, pellucid dots may be detected scattered irregularly through the leaf, sometimes having a faint yellowish color, which are the centers of infection. The appearance of the dots has a close resemblance to those of the oil glands in the leaves of the common St. John's wort (*Hypericum perforatum*), a rather abundant weed

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[1] Read at the meeting of the Amer. Ass'n Adv. Sci., Detroit, Mich., August 1897.

[2] J. C. ARTHUR, Proc. Amer. Ass'n Adv. Sci. **38** : 280. 1889.

[3] J. C. ARTHUR, Amer. Florist **6** : 419. 1891 ; Rep't Amer. Carnation Soc. 52. 1892; Rep't Amer. Carnation Soc. 12. 1894; Amer. Florist **9**: 467. 1894.

[4] ARTHUR and BOLLEY, Purdue Univ. Agr. Exp. Sta. Bull. no. 59. 1–38. *pl. 8.* 1896.

[5] The lily disease has been fully discussed in a bulletin soon to be issued by the Division of Vegetable Physiology and Pathology, U. S. Department of Agriculture.

or in the leaves of the false indigo (*Amorpha fruticosa*), a native shrub, except that they have no regular disposition. Sometimes the surface of the leaf is slightly raised over the dots, making watery pimples.

After a time the surface of the leaf above the dots changes enough to indicate their presence, and finally shows a distinct spot. As the disease extends inside the leaf the surface tissues collapse and whitish sunken spots appear. In some colored varieties of carnation the spots vary somewhat by being more or less reddish or purplish. As the spots increase in size the leaves wither, still clinging to the stem. Such spots never show distinct darker colored specks and rarely any concentric circles, as do the spots made by parasitic fungi, such as Septoria (spot disease) and Heterosporium (fairy ring).

Very badly diseased plants, especially when much crowded and growing in damp atmosphere, have more yellowish green leaves than normal, of a more transparent appearance, and usually smaller. The lower leaves of diseased plants in any atmosphere or soil die prematurely and the vitality of the plant is so lowered as to check the growth and decrease the size and number of the flowers.

Arthur and Bolley find that "no varieties of the carnation are exempt from the disease, but they differ much in susceptibility. The seat of this difference is chiefly in the vigor of the plant. . . . . Poorly grown plants are more affected than those well grown. Partly starved or stunted plants are specially liable to attack." They found the disease at Indianapolis, La Fayette, and many other places in Indiana, and in Buffalo, Boston, New York, Toronto, Chicago, and Lincoln. They say it is common throughout eastern North America wherever carnations are grown extensively, and conclude that it is caused by "parasitic bacteria entering the plant from the air through the stomata, or occasionally through the punctures of aphides."[6]

The disease which has formed the subject of our studies agrees in all essential points with the one just described, except that it is not caused by bacteria. Our material was obtained from many of the large centers of carnation growing in the United States, and through the kindness of Dr. Arthur good specimens were recently sent in from Mr. Fred Dorner's place at La Fayette, Ind.

[6] ARTHUR and BOLLEY, *loc. cit.* 32 and 37.

METHODS OF STUDY.

Microscopic investigations.—Material showing various stages of the disease was killed by both the chromic acid and absolute alcohol methods, dehydrated with alcohol, and infiltrated with paraffin in the usual way. Microtome sections were then cut and mounted in series, and these were stained with Ziel's carbol fuchsin, and, according to Gram's method, with anilin water gentian violet. The cells of the diseased spot were found to be much larger than normal, thin-walled, and oedemic. The chloroplasts were smaller than in healthy cells and were colorless or yellowish. Even after the most thorough and careful staining no parasitic or saprophytic organism could be detected in the tissues of these spots before the epidermal cells collapsed. In some cases after they collapsed fungi and bacteria were readily distinguished.

Cultures.—After many trials by washing and flaming it was found to be impossible to free the surface of a carnation leaf from outside germs by any means which might not also destroy germs in the tissues. Cultures were therefore made from the diseased mesophyll direct by very carefully peeling off the epidermis and scraping out the inner tissues with a flamed scraper which had been allowed to become perfectly cool. From one to twenty spots, varying in size from 0.5^{mm} to 2^{mm} in diameter, were included in each culture. Altogether about 500 cultures were made in various media, such as slightly acid, neutral, and slightly alkaline beef broth, with and without peptone; potato broth of various strengths; cauliflower broth; potato cylinders; agars of various composition; gelatin, acid, neutral, and alkaline to litmus, etc. In no case were organisms found in any culture made from a spot before the epidermal tissues had collapsed In cultures made from spots which had collapsed various fungi and bacteria were occasionally obtained, but not constantly or always of the same sort. Cultures including the epidermis frequently contained various organisms, among which was a yellow bacterium which occurred frequently on the surface of both diseased and healthy leaves, and which resembled in many respects the organism described by Arthur and Bolley as *Bacterium dianthi*, the cause of "bacteriosis." Infection experiments were made with this and other organisms, but the disease was not produced in any case.

From this work and the fact that no fungi or bacteria whatever were found associated with the disease in its earlier stages we were led to look in other directions for the cause.

RELATION OF APHIDES AND THRIPS TO THE DISEASE.

Aphides.—Early in the work our attention was called to the manner in which aphides attack the plant. A careful study revealed the fact that the insects in question were seldom if ever absent from plants, and that when present in limited numbers they were capable of producing effects identical in every way with those described under the name of "bacteriosis."

Studies of serial sections of spots produced by the aphides showed a breaking or laceration of the mesophyll cells, extending from the epidermis to various depths into the leaf. Oedemic swellings start from these points, resulting eventually in the development of all the characteristic symptoms described. Colonization experiments with the aphides were repeatedly made and it was found that the insects alone were capable of producing the disease and that neither fungi nor bacteria were present until the malady was well advanced, and in such cases not regularly nor constantly. In the colonization work it was found that the insects crawl between the young leaves on the growing shoots and do most of their work where they cannot be reached by tobacco smoke in sufficient strength to kill them. It was further found that it takes about two weeks for a puncture to become visible to the naked eye, appearing first as a minute translucent dot, accompanied by slight swellings of the tissues. Severe cases of the trouble were produced in perfectly healthy plants by colonizing the insects upon them, and the new growth of badly diseased plants was kept entirely free from the malady by eliminating the aphides and other puncturing insects. These results were obtained repeatedly in the laboratory and in the greenhouse.

Thrips.—Another form of spotting, usually accompanied by distortion of the young leaves and often of the stems, was found to be produced by thrips. These spots can be readily distinguished from the work of aphides, being very irregular in shape and having mossy outlines. The changes produced in the cells of the leaf, however, are apparently the same as those brought on by aphides, except that the mesophyll cells are not lacerated. Sections and cultures were repeatedly made from such spots produced under our control, but no organisms were obtained from the mesophyll in any case.

Throughout the work it was found that certain varieties of carnations were much more subject to the attacks of both aphides and thrips than others. Susceptibility was found to depend more upon the inherent characteristics of the individual plants than upon the variety. In most cases, however, susceptibility to attack is the direct result of improper methods of cultivation This is only expressing in another way a fact already well recognized by carnation growers, namely, that the disease under consideration is one which in most cases can be readily controlled by care in the selection and propagation of stock and attention to all the details of cultural conditions.

Summarizing it may be said that

1. The disease of carnations characterized by the symptoms already described and generally known as "bacteriosis" is wide spread and destructive.

2. In the earlier stages of the disease neither fungi nor bacteria are present, so far as can be determined by the most careful microscopic studies and bacteriological investigations.

3. As the disease advances various organisms appear, but their presence is not constant.

4. Infection experiments with such organisms, carried on under rigid bacteriological conditions, resulted negatively in every case.

5. A disease having all the characteristic symptoms of "bac-

teriosis," excepting the presence of bacteria, is produced by the puncture of aphides, as was repeatedly proved by the colonization of these insects on the plants.

6. That the aphides alone are responsible for the trouble is shown by the fact that the injuries produced are not accompanied in the earlier stages by fungi or bacteria. The aphides, therefore, can not be looked upon as carriers of any fungus or germ.

7. Injuries similar in many respects to those produced by aphides also result from the attacks of thrips, an insect nearly always present on carnations under glass, although generally overlooked by growers.

8. The carnation is a plant readily influenced by the conditions under which it is grown, and as a result the reaction to the injury of the aphides and the susceptibility to their attacks not only vary with varieties, but with individuals of the same variety. Plants, therefore, grown under improper conditions will show more of the characteristic injuries from a given number of aphis punctures than those where all the conditions have been favorable for growth.

9. Proper selection and propagation of stock; furnishing soil, moisture, light, and air best adapted to healthy and vigorous growth; and keeping down to a minimum the number of aphides and thrips will enable the grower to successfully combat the disease.

DIVISION OF VEGETABLE PHYSIOLOGY AND PATHOLOGY,
U. S. DEPARTMENT OF AGRICULTURE.